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| NEWS 13 | DEC 01 | DGENE, USGENE, and PCTGEN: new percent identity feature for sorting BLAST answer sets |
| NEWS 14 | DEC 02 | Derwent World Patent Index: Japanese FI-TERM thesaurus added |
| NEWS 15 | DEC 02 | PCTGEN enhanced with patent family and legal status display data from INPADOCDB |
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=> s thrombopoietin
L1       28224 THROMBOPOIETIN

=> s l1 and human
   6 FILES SEARCHED...
L2       17018 L1 AND HUMAN

=> s  L2 and purification
L3        1051 L2 AND PURIFICATION

=> s thrombopoietin purification
L4           7 THROMBOPOIETIN PURIFICATION

=> s l3 and l4
L5           7 L3 AND L4

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L4   ANSWER 1 OF 7  WPIDS COPYRIGHT 2009       THOMSON REUTERS on STN
TI   Thrombopoietin purification by removal of protein
     contaminants using hydroxyapatite  - provides homogenous preparation of
     thrombopoietin substantially free of contaminants
AN   1997-052235 [05]    WPIDS
AB   WO 1996040773 A1   UPAB: 20060112
     A novel method for purifying thrombopoietin (TPO) from a biological fluid
     comprises: (a) reducing the volume of a TPO-containing biological fluid by a
     method selected from the gp. consisting of ligand affinity chromatography,
```

ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concentration of the concentrated fraction to provide an adjusted solution; (c) acidifying the adjusted solution to precipitate contaminant proteins and provide a cleared solution; (d) fractionating the cleared solution by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a solution comprising TPO and protein contaminants, comprising exposing the solution to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepared by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0003)
ABEQ EP 839158 A1 UPAB 20060112

A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0004)
ABEQ US 5744587 A UPAB 20060112

A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln.

comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0007)

ABEQ JP 11507033 W UPAB 20060112

A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

ACCESSION NUMBER: 1997-052235 [05] WPIDS
DOC. NO. CPI: C1997-017386 [05]
TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants
DERWENT CLASS: A96; B04; D16
INVENTOR: ALASKA A A; ALASKA A R; CHANG J; DOWNEY W; FORSTROM J W; PHAN L; ALASKA A; FORSTROM W
PATENT ASSIGNEE: (ZYMO-C) ZYMOGENETICS INC
COUNTRY COUNT: 69

PATENT INFO ABBR.:

| PATENT NO | KIND | DATE | WEEK | LA | PG | MAIN IPC |
|---|---|---|---|---|---|---|
| WO 9640773 | A1 | 19961219 | (199705)* | EN | 33[0] | |
| AU 9658718 | A | 19961230 | (199716) | EN | | |
| EP 839158 | A1 | 19980506 | (199822) | EN | [0] | |
| US 5744587 | A | 19980428 | (199824) | EN | 9[0] | |
| AU 694043 | B | 19980709 | (199838) | EN | | |
| NZ 308862 | A | 19990128 | (199910) | EN | | |
| JP 11507033 | W | 19990622 | (199935) | JA | 29 | |
| MX 9709312 | A1 | 19980201 | (199954) | ES | | |
| KR 99022541 | A | 19990325 | (200023) | KO | [0] | |
| CA 2223236 | C | 20000919 | (200054) | EN | | |
| KR 255466 | B1 | 20000501 | (200128) | KO | | |
| MX 205114 | B | 20011109 | (200279) | ES | | |

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|---|---|---|---|
| CN 1187202 | A | 19980708 (200336) | ZH |
| EP 839158 | B1 | 20051228 (200605) | EN |
| DE 69635661 | E | 20060202 (200615) | DE |
| DE 69635661 | T2 | 20060720 (200652) | DE |

APPLICATION DETAILS:

| PATENT NO | KIND | APPLICATION DATE |
|---|---|---|
| WO 9640773 A1 | | WO 1996-US7453 19960522 |
| US 5744587 A | | US 1995-484246 19950607 |
| AU 9658718 A | | AU 1996-58718 19960522 |
| AU 694043 B | | AU 1996-58718 19960522 |
| CA 2223236 C | | CA 1996-2223236 19960522 |
| CN 1187202 A | | CN 1996-194563 19960522 |
| DE 69635661 E | | DE 1996-635661 19960522 |
| EP 839158 A1 | | EP 1996-920393 19960522 |
| EP 839158 B1 | | EP 1996-920393 19960522 |
| DE 69635661 E | | EP 1996-920393 19960522 |
| NZ 308862 A | | NZ 1996-308862 19960522 |
| EP 839158 A1 | | WO 1996-US7453 19960522 |
| NZ 308862 A | | WO 1996-US7453 19960522 |
| JP 11507033 W | | WO 1996-US7453 19960522 |
| KR 99022541 A | | WO 1996-US7453 19960522 |
| CA 2223236 C | | WO 1996-US7453 19960522 |
| KR 255466 B1 | | WO 1996-US7453 19960522 |
| CN 1187202 A | | WO 1996-US7453 19960522 |
| EP 839158 B1 | | WO 1996-US7453 19960522 |
| DE 69635661 E | | WO 1996-US7453 19960522 |
| JP 11507033 W | | JP 1997-500670 19960522 |
| MX 9709312 A1 | | MX 1997-9312 19971201 |
| MX 205114 B | | MX 1997-9312 19971201 |
| KR 99022541 A | | KR 1997-709022 19971206 |
| KR 255466 B1 | | KR 1997-709022 19971206 |
| DE 69635661 T2 | | DE 1996-635661 19960522 |
| DE 69635661 T2 | | EP 1996-920393 19960522 |
| DE 69635661 T2 | | WO 1996-US7453 19960522 |

FILING DETAILS:

| PATENT NO | KIND | | PATENT NO | |
|---|---|---|---|---|
| AU 694043 | B | Previous Publ | AU 9658718 | A |
| DE 69635661 | E | Based on | EP 839158 | A |
| AU 9658718 | A | Based on | WO 9640773 | A |
| EP 839158 | A1 | Based on | WO 9640773 | A |
| AU 694043 | B | Based on | WO 9640773 | A |
| NZ 308862 | A | Based on | WO 9640773 | A |
| JP 11507033 | W | Based on | WO 9640773 | A |
| KR 99022541 | A | Based on | WO 9640773 | A |
| CA 2223236 | C | Based on | WO 9640773 | A |
| CN 1187202 | A | Based on | WO 9640773 | A |
| EP 839158 | B1 | Based on | WO 9640773 | A |
| DE 69635661 | E | Based on | WO 9640773 | A |
| DE 69635661 | T2 | Based on | EP 839158 | A |
| DE 69635661 | T2 | Based on | WO 9640773 | A |

PRIORITY APPLN. INFO: US 1995-484246 19950607
WO 1996-US7453 19960522

L4 ANSWER 2 OF 7 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN

TI Thrombopoietin purification by removal of protein contaminants using hydroxyapatite;
human thrombopoietin purification using hydroxyapatite chromatography
AN 1997-01852 BIOTECHDS
AB A method for purifying human thrombopoietin (TPO) from a biological fluid is claimed, and involves: (1) reducing the column of a TPO-containing fluid by ligand affinity chromatography, ionexchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (2) adjusting the salt concentration of the concentration fraction; (3) acidifying the adjusted solution to precipitate contaminant proteins and provide a cleared solution; (4) fractionating the cleared solution by anion-exchange chromatography to provide a TPO-enriched fraction; (5) exposing this fraction to hydroxyapatite chromatography so that protein contaminants remain bound to the column and the TPO remains unbound; (6) collecting the unbound TPO; and (7) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. The biological fluid is a cell-conditioned culture medium. TPO obtained by this method can be used therapeutically e.g. in the treatment of cytopenias. (33pp)
ACCESSION NUMBER: 1997-01852 BIOTECHDS
TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite;
human thrombopoietin purification using hydroxyapatite chromatography
AUTHOR: Alaska A R; Chang J J; Downey W; Forstrom J W; Phan L
PATENT ASSIGNEE: Zymogenetics
LOCATION: Seattle, WA, USA.
PATENT INFO: WO 9640773 19 Dec 1996
APPLICATION INFO: WO 1996-US7453 22 May 1996
PRIORITY INFO: US 1995-484246 7 Jun 1995
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-052235 [05]

L4 ANSWER 3 OF 7 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI New pure thrombopoietin free of low-mol.weight degradation products;
purification from cell culture supernatant or milk by MPL receptor ligand-binding domain affinity chromatography and anion-exchange chromatography
AN 1996-11056 BIOTECHDS
AB A new purified mammal thrombopoietin (TPO) has a mol.weight of 70,000 +/- 10,000 (denaturing SDS-PAGE), is at least 90% pure as determined by SDS-PAGE and silver staining, and is free of TPO species of mol.weight less than 55,000. The TPO may be of mouse, primate or human origin, with a specified protein sequence. The TPO is purified from a conditioned cell culture supernatant or milk by an optional concentration step, affinity chromatography against a ligand-binding domain of an MPL receptor on crosslinked agarose beads, and anion-exchange chromatography. TPO stimulates megakaryocytopoiesis and thrombocytopoiesis, and may be used to increase the level of platelets in the blood, e.g. in cases of aplastic anemia, myelodysplastic syndrome, chemotherapy, congenital cytopenia, etc., and may also be used to increase the number of circulating erythrocytes (or precursors), especially in therapy of anemia associated with bone marrow failure. The new TPO preparation is homogeneous and free of proteolytic degradation products. Its use reduces the need for transfusion and thus the risk of platelet alloimmunity. (91pp)
ACCESSION NUMBER: 1996-11056 BIOTECHDS
TITLE: New pure thrombopoietin free of low-mol.weight degradation products;

purification from cell culture supernatant or milk by MPL receptor ligand-binding domain affinity chromatography and anion-exchange chromatography
AUTHOR: Forstrom J W; Lofton-Day C E; Lok S
PATENT ASSIGNEE: Zymogenetics
LOCATION: Seattle, WA, USA.
PATENT INFO: WO 9620955 11 Jul 1996
APPLICATION INFO: WO 1995-US16626 20 Dec 1995
PRIORITY INFO: US 1994-366859 30 Dec 1994
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1996-333942 [33]

L4 ANSWER 4 OF 7 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI Hematopoietic proteins and polypeptides;
useful in in vivo and ex vivo therapy
AN 1995-13081 BIOTECHDS
AB A mouse or human hematopoietic protein (I) (protein sequences disclosed) stimulating the proliferation or differentiation of myeloid or lymphoid precursors is claimed. Also claimed are: proteins with at least 80% homology to (I); DNA encoding (I) (DNA sequence disclosed); DNA encoding (I), allelic variants, complementary sequences and DNA with at least 80% homology to the DNA encoding (I); the EcoRI-XhoI insert of plasmid pZGmp1-1081 (ATCC 69566) and its allelic variants; an expression vector containing a transcription promoter and a (I)-encoding DNA segment; a transformed fungus, yeast, bacterium or mammal cell culture containing the vector; a non-human transgenic animal containing the claimed DNA sequences in its germline; production of recombinant hematopoietic protein by culturing the transformed cell culture; a pharmaceutical composition of (I); an antibody; a method for stimulating platelet production in a mammal using (I); a DNA probe; a method for detecting DNA encoding thrombopoietin using the DNA probe; a method for stimulating cell proliferation using (I); and a method for thrombopoietin purification using the antibody. (137pp)
ACCESSION NUMBER: 1995-13081 BIOTECHDS
TITLE: Hematopoietic proteins and polypeptides;
useful in in vivo and ex vivo therapy
AUTHOR: Holly R D; Lok S; Foster D C; Hagen F S; Kaushansky K; Kuijper J L; Lofton-Day C; Oort P J; Burkhead S K
PATENT ASSIGNEE: Zymogenetics; Univ.Washington-Seattle
PATENT INFO: WO 9521920 17 Aug 1995
APPLICATION INFO: WO 1994-US8806 5 Aug 1994
PRIORITY INFO: US 1994-525491 1 Jun 1994; US 1994-196025 14 Feb 1994
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1995-293121 [38]

L4 ANSWER 5 OF 7 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI Studies on the purification of thrombopoietin from kidney cell culture protein;
using ammonium sulfate fractionation and chromatography
AN 1985-10996 BIOTECHDS
AB A thrombocytopoiesis-stimulating factor (TSF) has been purified from human embryonic kidney (HEK) cell culture medium. In the initial purification step, crude HEK cell culture medium was fractionated with saturated ammonium sulfate. The proteins precipitated at 40-60% and 60-80% saturation increased the % of sulfur 35 incorporation into platelets of assay mice. These proteins were refined on Sephadex G-75 columns, and the fraction containing the highest specific activity was purified by DEAE-cellulose column chromatography. TSF activity was eluted from the columns between 0.3 and 1.0 mol/l NaCl. Additional Sephadex

chromatography of post-DEAE-chromatographic preparations further increased the purity of the TSF. TSF was further processed on a DEAE HPLC column or size exclusion (SE)-HPLC columns. After HPLC, the activity was localized in a region corresponding to a retention time of 6 to 8 min for the DEAE-HPLC, but longer times were found after SE-HPLC. TSF was further purified by additional SDS-PAGE and SE-HPLC. The final product had significant TSF activity and represented a purification of about 500,000-fold. (22 ref)

ACCESSION NUMBER: 1985-10996 BIOTECHDS
TITLE: Studies on the purification of thrombopoietin from kidney cell culture protein; using ammonium sulfate fractionation and chromatography
AUTHOR: McDonald T P; Cottrell M; Clift R; Khouri J A; Long M D
CORPORATE SOURCE: Abbott
LOCATION: University of Tennessee College of Veterinary Medicine, P.O. Box 1071, Knoxville, TN 37901-1071, USA.
SOURCE: J.Lab.Clin.Med.; (1985) 106, 2, 162-74
CODEN: JLCMAK
DOCUMENT TYPE: Journal
LANGUAGE: English

L4 ANSWER 6 OF 7 DGENE COPYRIGHT 2009 THOMSON REUTERS on STN
TI Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants
AN AAW22466 protein DGENE
AB AAW22465 and AAW22466 represent the mouse and human thrombopoietins (TPO), respectively. These sequences can be purified using the method of the invention. The method of the invention is for purifying TPO from a biological fluid. The method comprises reducing the volume of a TPO containing biological fluid to provide a concentrated fraction, adjusting the salt concentration of the fraction, and acidifying the adjusted solution to precipitate contaminant proteins. The cleared solution is fractionated to give a TPO enriched fraction. The TPO-enriched fraction is exposed to hydroxyapatite, and TPO remains substantially unbound while contaminants bind to the hydroxyapatite. The unbound TPO is collected and concentrated. TPO prepared by this method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.
ACCESSION NUMBER: AAW22466 protein DGENE
TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants
INVENTOR: Alaska A R; Chang J; Downey W; Forstrom J W; Phan L
PATENT ASSIGNEE: (ZYMO)ZYMOGENETICS INC.
PATENT INFO: WO 9640773 A1 19961219 33
APPLICATION INFO: WO 1996-US7453 19960522
PRIORITY INFO: US 1995-484246 19950607
PAT. SEQ. LOC: Disclosure; Page 23-25
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: 1997-052235 [05]
DESCRIPTION: Human thrombopoietin.

L4 ANSWER 7 OF 7 DGENE COPYRIGHT 2009 THOMSON REUTERS on STN
TI Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants

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AN    AAW22465  protein       DGENE
AB    AAW22465 and AAW22466 represent the mouse and human thrombopoietins
      (TPO), respectively. These sequences can be purified using the method of
      the invention. The method of the invention is for purifying TPO from a
      biological fluid. The method comprises reducing the volume of a TPO
      containing biological fluid to provide a concentrated fraction, adjusting
      the salt concentration of the fraction, and acidifying the adjusted
      solution to precipitate contaminant proteins. The cleared solution is
      fractionated to give a TPO enriched fraction. The TPO-enriched fraction
      is exposed to hydroxyapatite, and TPO remains substantially unbound while
      contaminants bind to the hydroxyapatite. The unbound TPO is collected and
      concentrated. TPO prepared by this method can be used therapeutically
      wherever it is desirable to increase proliferation of cells in the bone
      marrow, such as in the treatment of cytopenias, e.g. induced by aplastic
      anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias
      Revised record issued on 15-JUN-2007 : Enhanced with precomputed
      information from BOND.
ACCESSION NUMBER: AAW22465  protein       DGENE
TITLE:            Thrombopoietin purification by removal of
                  protein contaminants using hydroxyapatite  - provides
                  homogenous preparation of thrombopoietin substantially free
                  of contaminants
INVENTOR:         Alaska A R; Chang J; Downey W; Forstrom J W; Phan L
PATENT ASSIGNEE:  (ZYMO)ZYMOGENETICS INC.
PATENT INFO:      WO 9640773      A1 19961219                33
APPLICATION INFO: WO 1996-US7453       19960522
PRIORITY INFO:    US 1995-484246       19950607
PAT. SEQ. LOC:    Disclosure; Page 22-23
DOCUMENT TYPE:    Patent
LANGUAGE:         English
OTHER SOURCE:     1997-052235 [05]
CROSS REFERENCES: PC-NCBI: gi506827
                  PC-SWISSPROT: P40225
DESCRIPTION:      Mouse thrombopoietin.


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     FILE 'MEDLINE, BIOSIS, WPIDS, HCAPLUS, BIOTECHDS, DGENE, EMBASE,
     SCISEARCH' ENTERED AT 18:05:53 ON 11 DEC 2009
L1        28224 S THROMBOPOIETIN
L2        17018 S L1 AND HUMAN
L3         1051 S  L2 AND PURIFICATION
L4            7 S THROMBOPOIETIN PURIFICATION
L5            7 S L3 AND L4

=> s l3 and (affinity chromatography)
L6          17 L3 AND (AFFINITY CHROMATOGRAPHY)

=> s l6 and (reverse phase chromatography)
L7           0 L6 AND (REVERSE PHASE CHROMATOGRAPHY)

=> s l6 and (hydrophobic interaction chromatography)
L8           2 L6 AND (HYDROPHOBIC INTERACTION CHROMATOGRAPHY)

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L8   ANSWER 1 OF 2  WPIDS COPYRIGHT 2009       THOMSON REUTERS on STN
TI   Thrombopoietin purification by removal of protein
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contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants

AN 1997-052235 [05] WPIDS

AB WO 1996040773 A1 UPAB: 20060112
A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-containing biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concentration of the concentrated fraction to provide an adjusted solution; (c) acidifying the adjusted solution to precipitate contaminant proteins and provide a cleared solution; (d) fractionating the cleared solution by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a solution comprising TPO and protein contaminants, comprising exposing the solution to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepared by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0003)

ABEQ EP 839158 A1 UPAB 20060112
A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0004)

ABEQ US 5744587 A UPAB 20060112
A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and

ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0007)

ABEQ JP 11507033 W UPAB 20060112

A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

ACCESSION NUMBER: 1997-052235 [05] WPIDS

DOC. NO. CPI: C1997-017386 [05]

TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants

DERWENT CLASS: A96; B04; D16

INVENTOR: ALASKA A A; ALASKA A R; CHANG J; DOWNEY W; FORSTROM J W; PHAN L; ALASKA A; FORSTROM W

PATENT ASSIGNEE: (ZYMO-C) ZYMOGENETICS INC

COUNTRY COUNT: 69

PATENT INFO ABBR.:

| PATENT NO | KIND DATE | WEEK | LA | PG | MAIN IPC |
|---|---|---|---|---|---|
| WO 9640773 | A1 19961219 | (199705)* | EN | 33[0] | |

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| AU 9658718 | A | 19961230 (199716) | EN | |
| EP 839158 | A1 | 19980506 (199822) | EN | [0] |
| US 5744587 | A | 19980428 (199824) | EN | 9[0] |
| AU 694043 | B | 19980709 (199838) | EN | |
| NZ 308862 | A | 19990128 (199910) | EN | |
| JP 11507033 | W | 19990622 (199935) | JA | 29 |
| MX 9709312 | A1 | 19980201 (199954) | ES | |
| KR 99022541 | A | 19990325 (200023) | KO | [0] |
| CA 2223236 | C | 20000919 (200054) | EN | |
| KR 255466 | B1 | 20000501 (200128) | KO | |
| MX 205114 | B | 20011109 (200279) | ES | |
| CN 1187202 | A | 19980708 (200336) | ZH | |
| EP 839158 | B1 | 20051228 (200605) | EN | |
| DE 69635661 | E | 20060202 (200615) | DE | |
| DE 69635661 | T2 | 20060720 (200652) | DE | |

APPLICATION DETAILS:

| PATENT NO | KIND | APPLICATION DATE |
|---|---|---|
| WO 9640773 A1 | | WO 1996-US7453 19960522 |
| US 5744587 A | | US 1995-484246 19950607 |
| AU 9658718 A | | AU 1996-58718 19960522 |
| AU 694043 B | | AU 1996-58718 19960522 |
| CA 2223236 C | | CA 1996-2223236 19960522 |
| CN 1187202 A | | CN 1996-194563 19960522 |
| DE 69635661 E | | DE 1996-635661 19960522 |
| EP 839158 A1 | | EP 1996-920393 19960522 |
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| DE 69635661 E | | EP 1996-920393 19960522 |
| NZ 308862 A | | NZ 1996-308862 19960522 |
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| NZ 308862 A | | WO 1996-US7453 19960522 |
| JP 11507033 W | | WO 1996-US7453 19960522 |
| KR 99022541 A | | WO 1996-US7453 19960522 |
| CA 2223236 C | | WO 1996-US7453 19960522 |
| KR 255466 B1 | | WO 1996-US7453 19960522 |
| CN 1187202 A | | WO 1996-US7453 19960522 |
| EP 839158 B1 | | WO 1996-US7453 19960522 |
| DE 69635661 E | | WO 1996-US7453 19960522 |
| JP 11507033 W | | JP 1997-500670 19960522 |
| MX 9709312 A1 | | MX 1997-9312 19971201 |
| MX 205114 B | | MX 1997-9312 19971201 |
| KR 99022541 A | | KR 1997-709022 19971206 |
| KR 255466 B1 | | KR 1997-709022 19971206 |
| DE 69635661 T2 | | DE 1996-635661 19960522 |
| DE 69635661 T2 | | EP 1996-920393 19960522 |
| DE 69635661 T2 | | WO 1996-US7453 19960522 |

FILING DETAILS:

| PATENT NO | KIND | | PATENT NO | |
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| AU 694043 | B | Previous Publ | AU 9658718 | A |
| DE 69635661 | E | Based on | EP 839158 | A |
| AU 9658718 | A | Based on | WO 9640773 | A |
| EP 839158 | A1 | Based on | WO 9640773 | A |
| AU 694043 | B | Based on | WO 9640773 | A |
| NZ 308862 | A | Based on | WO 9640773 | A |
| JP 11507033 | W | Based on | WO 9640773 | A |
| KR 99022541 | A | Based on | WO 9640773 | A |

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|---|---|---|---|---|---|
| CA 2223236 | C | Based on | WO 9640773 | A |
| CN 1187202 | A | Based on | WO 9640773 | A |
| EP 839158 | B1 | Based on | WO 9640773 | A |
| DE 69635661 | E | Based on | WO 9640773 | A |
| DE 69635661 | T2 | Based on | EP 839158 | A |
| DE 69635661 | T2 | Based on | WO 9640773 | A |

PRIORITY APPLN. INFO: US 1995-484246 19950607
WO 1996-US7453 19960522

L8 ANSWER 2 OF 2 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI Thrombopoietin purification by removal of protein contaminants using hydroxyapatite; human thrombopoietin purification using hydroxyapatite chromatography
AN 1997-01852 BIOTECHDS
AB A method for purifying human thrombopoietin (TPO) from a biological fluid is claimed, and involves: (1) reducing the column of a TPO-containing fluid by ligand affinity chromatography, ionexchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (2) adjusting the salt concentration of the concentration fraction; (3) acidifying the adjusted solution to precipitate contaminant proteins and provide a cleared solution; (4) fractionating the cleared solution by anion-exchange chromatography to provide a TPO-enriched fraction; (5) exposing this fraction to hydroxyapatite chromatography so that protein contaminants remain bound to the column and the TPO remains unbound; (6) collecting the unbound TPO; and (7) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. The biological fluid is a cell-conditioned culture medium. TPO obtained by this method can be used therapeutically e.g. in the treatment of cytopenias. (33pp)
ACCESSION NUMBER: 1997-01852 BIOTECHDS
TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite; human thrombopoietin purification using hydroxyapatite chromatography
AUTHOR: Alaska A R; Chang J J; Downey W; Forstrom J W; Phan L
PATENT ASSIGNEE: Zymogenetics
LOCATION: Seattle, WA, USA.
PATENT INFO: WO 9640773 19 Dec 1996
APPLICATION INFO: WO 1996-US7453 22 May 1996
PRIORITY INFO: US 1995-484246 7 Jun 1995
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-052235 [05]

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(FILE 'HOME' ENTERED AT 18:04:37 ON 11 DEC 2009)

FILE 'MEDLINE, BIOSIS, WPIDS, HCAPLUS, BIOTECHDS, DGENE, EMBASE, SCISEARCH' ENTERED AT 18:05:53 ON 11 DEC 2009

| | | | |
|---|---|---|---|
| L1 | 28224 | S | THROMBOPOIETIN |
| L2 | 17018 | S | L1 AND HUMAN |
| L3 | 1051 | S | L2 AND PURIFICATION |
| L4 | 7 | S | THROMBOPOIETIN PURIFICATION |
| L5 | 7 | S | L3 AND L4 |
| L6 | 17 | S | L3 AND (AFFINITY CHROMATOGRAPHY) |
| L7 | 0 | S | L6 AND (REVERSE PHASE CHROMATOGRAPHY) |

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L8             2 S L6 AND (HYDROPHOBIC INTERACTION CHROMATOGRAPHY)

=> s l6 and (anion exchange chromatography)
L9             3 L6 AND (ANION EXCHANGE CHROMATOGRAPHY)

=> d l9 ti abs ibib tot

L9   ANSWER 1 OF 3  WPIDS COPYRIGHT 2009       THOMSON REUTERS on STN
TI   Thrombopoietin purification by removal of protein
     contaminants using hydroxyapatite  - provides homogenous preparation of
     thrombopoietin substantially free of contaminants
AN   1997-052235 [05]   WPIDS
AB   WO 1996040773 A1   UPAB: 20060112
     A novel method for purifying thrombopoietin (TPO) from a
     biological fluid comprises: (a) reducing the volume of a TPO-containing
     biological fluid by a method selected from the gp. consisting of ligand
     affinity chromatography, ion-exchange chromatography,
     hydrophobic interaction chromatography and ultrafiltration to provide a
     concentrated fraction; (b) adjusting the salt concentration of the concentrated
     fraction to provide an adjusted solution; (c) acidifying the adjusted solution
     to precipitate contaminant proteins and provide a cleared solution; (d)
     fractionating the cleared solution by anion-exchange
     chromatography to provide a TPO-enriched fraction; (e) exposing
     the TPO-enriched fraction to hydroxyapatite where protein contaminants are
     bound to the hydroxyapatite and TPO remains substantially unbound; (f)
     collecting the unbound TPO; and (g) concentrating the collected TPO by
     cation-exchange chromatography or ultrafiltration. Also claimed is a
     method for removing protein contaminants from a solution comprising TPO and
     protein contaminants, comprising exposing the solution to hydroxyapatite,
     where the protein contaminants are bound to the hydroxyapatite and the TPO
     remains substantially unbound, and recovering the TPO.
          USE - TPO prepared by the above method can be used therapeutically
     wherever it is desirable to increase proliferation of cells in the bone
     marrow, such as in the treatment of cytopenias, e.g. induced by aplastic
     anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0003)
ABEQ EP 839158 A1   UPAB 20060112
     A novel method for purifying thrombopoietin (TPO) from a
     biological fluid comprises: (a) reducing the volume of a TPO-contg.
     biological fluid by a method selected from the gp. consisting of ligand
     affinity chromatography, ion-exchange chromatography,
     hydrophobic interaction chromatography and ultrafiltration to provide a
     concentrated fraction; (b) adjusting the salt concn. of the concentrated
     fraction to provide an adjusted soln.; (c) acidifying the adjusted soln.
     to precipitate contaminant proteins and provide a cleared soln.; (d)
     fractionating the cleared soln. by anion-exchange
     chromatography to provide a TPO-enriched fraction; (e) exposing
     the TPO-enriched fraction to hydroxyapatite where protein contaminants are
     bound to the hydroxyapatite and TPO remains substantially unbound; (f)
     collecting the unbound TPO; and (g) concentrating the collected TPO by
     cation-exchange chromatography or ultrafiltration. Also claimed is a
     method for removing protein contaminants from a soln. comprising TPO and
     protein contaminants, comprising exposing the soln. to hydroxyapatite,
     where the protein contaminants are bound to the hydroxyapatite and the TPO
     remains substantially unbound, and recovering the TPO.
          USE - TPO prepd. by the above method can be used therapeutically
     wherever it is desirable to increase proliferation of cells in the bone
     marrow, such as in the treatment of cytopenias, e.g. induced by aplastic
     anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.
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Member(0004)
ABEQ US 5744587 A UPAB 20060112
A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

Member(0007)
ABEQ JP 11507033 W UPAB 20060112
A novel method for purifying thrombopoietin (TPO) from a biological fluid comprises: (a) reducing the volume of a TPO-contg. biological fluid by a method selected from the gp. consisting of ligand affinity chromatography, ion-exchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (b) adjusting the salt concn. of the concentrated fraction to provide an adjusted soln.; (c) acidifying the adjusted soln. to precipitate contaminant proteins and provide a cleared soln.; (d) fractionating the cleared soln. by anion-exchange chromatography to provide a TPO-enriched fraction; (e) exposing the TPO-enriched fraction to hydroxyapatite where protein contaminants are bound to the hydroxyapatite and TPO remains substantially unbound; (f) collecting the unbound TPO; and (g) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. Also claimed is a method for removing protein contaminants from a soln. comprising TPO and protein contaminants, comprising exposing the soln. to hydroxyapatite, where the protein contaminants are bound to the hydroxyapatite and the TPO remains substantially unbound, and recovering the TPO.

USE - TPO prepd. by the above method can be used therapeutically wherever it is desirable to increase proliferation of cells in the bone marrow, such as in the treatment of cytopenias, e.g. induced by aplastic anaemia, myelodisplastic syndromes, chemotherapy or congenital cytopenias.

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|---|---|
| ACCESSION NUMBER: | 1997-052235 [05] WPIDS |
| DOC. NO. CPI: | C1997-017386 [05] |
| TITLE: | Thrombopoietin purification by removal of protein contaminants using hydroxyapatite - provides homogenous preparation of thrombopoietin substantially free of contaminants |
| DERWENT CLASS: | A96; B04; D16 |
| INVENTOR: | ALASKA A A; ALASKA A R; CHANG J; DOWNEY W; FORSTROM J W; PHAN L; ALASKA A; FORSTROM W |
| PATENT ASSIGNEE: | (ZYMO-C) ZYMOGENETICS INC |
| COUNTRY COUNT: | 69 |

PATENT INFO ABBR.:

| PATENT NO | KIND | DATE | WEEK | LA | PG | MAIN IPC |
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| WO 9640773 | A1 | 19961219 | (199705)* | EN | 33[0] | |
| AU 9658718 | A | 19961230 | (199716) | EN | | |
| EP 839158 | A1 | 19980506 | (199822) | EN | [0] | |
| US 5744587 | A | 19980428 | (199824) | EN | 9[0] | |
| AU 694043 | B | 19980709 | (199838) | EN | | |
| NZ 308862 | A | 19990128 | (199910) | EN | | |
| JP 11507033 | W | 19990622 | (199935) | JA | 29 | |
| MX 9709312 | A1 | 19980201 | (199954) | ES | | |
| KR 99022541 | A | 19990325 | (200023) | KO | [0] | |
| CA 2223236 | C | 20000919 | (200054) | EN | | |
| KR 255466 | B1 | 20000501 | (200128) | KO | | |
| MX 205114 | B | 20011109 | (200279) | ES | | |
| CN 1187202 | A | 19980708 | (200336) | ZH | | |
| EP 839158 | B1 | 20051228 | (200605) | EN | | |
| DE 69635661 | E | 20060202 | (200615) | DE | | |
| DE 69635661 | T2 | 20060720 | (200652) | DE | | |

APPLICATION DETAILS:

| PATENT NO | KIND | APPLICATION | DATE |
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| WO 9640773 A1 | | WO | 1996-US7453 19960522 |
| US 5744587 A | | US | 1995-484246 19950607 |
| AU 9658718 A | | AU | 1996-58718 19960522 |
| AU 694043 B | | AU | 1996-58718 19960522 |
| CA 2223236 C | | CA | 1996-2223236 19960522 |
| CN 1187202 A | | CN | 1996-194563 19960522 |
| DE 69635661 E | | DE | 1996-635661 19960522 |
| EP 839158 A1 | | EP | 1996-920393 19960522 |
| EP 839158 B1 | | EP | 1996-920393 19960522 |
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| NZ 308862 A | | NZ | 1996-308862 19960522 |
| EP 839158 A1 | | WO | 1996-US7453 19960522 |
| NZ 308862 A | | WO | 1996-US7453 19960522 |
| JP 11507033 W | | WO | 1996-US7453 19960522 |
| KR 99022541 A | | WO | 1996-US7453 19960522 |
| CA 2223236 C | | WO | 1996-US7453 19960522 |
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| EP 839158 B1 | | WO | 1996-US7453 19960522 |
| DE 69635661 E | | WO | 1996-US7453 19960522 |
| JP 11507033 W | | JP | 1997-500670 19960522 |
| MX 9709312 A1 | | MX | 1997-9312 19971201 |
| MX 205114 B | | MX | 1997-9312 19971201 |
| KR 99022541 A | | KR | 1997-709022 19971206 |
| KR 255466 B1 | | KR | 1997-709022 19971206 |
| DE 69635661 T2 | | DE | 1996-635661 19960522 |
| DE 69635661 T2 | | EP | 1996-920393 19960522 |
| DE 69635661 T2 | | WO | 1996-US7453 19960522 |

FILING DETAILS:

| PATENT NO | KIND | | PATENT NO | |
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| AU 694043 | B | Previous Publ | AU 9658718 | A |
| DE 69635661 | E | Based on | EP 839158 | A |
| AU 9658718 | A | Based on | WO 9640773 | A |

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|---|---|---|---|---|---|
| EP 839158 | A1 | Based on | WO 9640773 | A |
| AU 694043 | B | Based on | WO 9640773 | A |
| NZ 308862 | A | Based on | WO 9640773 | A |
| JP 11507033 | W | Based on | WO 9640773 | A |
| KR 99022541 | A | Based on | WO 9640773 | A |
| CA 2223236 | C | Based on | WO 9640773 | A |
| CN 1187202 | A | Based on | WO 9640773 | A |
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| DE 69635661 | T2 | Based on | EP 839158 | A |
| DE 69635661 | T2 | Based on | WO 9640773 | A |

PRIORITY APPLN. INFO: US 1995-484246 19950607
WO 1996-US7453 19960522

L9 ANSWER 2 OF 3 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI Thrombopoietin purification by removal of protein contaminants using hydroxyapatite;
human thrombopoietin purification using hydroxyapatite chromatography
AN 1997-01852 BIOTECHDS
AB A method for purifying human thrombopoietin (TPO) from a biological fluid is claimed, and involves: (1) reducing the column of a TPO-containing fluid by ligand affinity chromatography, ionexchange chromatography, hydrophobic interaction chromatography and ultrafiltration to provide a concentrated fraction; (2) adjusting the salt concentration of the concentration fraction; (3) acidifying the adjusted solution to precipitate contaminant proteins and provide a cleared solution; (4) fractionating the cleared solution by anion-exchange chromatography to provide a TPO-enriched fraction; (5) exposing this fraction to hydroxyapatite chromatography so that protein contaminants remain bound to the column and the TPO remains unbound; (6) collecting the unbound TPO; and (7) concentrating the collected TPO by cation-exchange chromatography or ultrafiltration. The biological fluid is a cell-conditioned culture medium. TPO obtained by this method can be used therapeutically e.g. in the treatment of cytopenias. (33pp)
ACCESSION NUMBER: 1997-01852 BIOTECHDS
TITLE: Thrombopoietin purification by removal of protein contaminants using hydroxyapatite; human thrombopoietin purification using hydroxyapatite chromatography
AUTHOR: Alaska A R; Chang J J; Downey W; Forstrom J W; Phan L
PATENT ASSIGNEE: Zymogenetics
LOCATION: Seattle, WA, USA.
PATENT INFO: WO 9640773 19 Dec 1996
APPLICATION INFO: WO 1996-US7453 22 May 1996
PRIORITY INFO: US 1995-484246 7 Jun 1995
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1997-052235 [05]

L9 ANSWER 3 OF 3 BIOTECHDS COPYRIGHT 2009 THOMSON REUTERS on STN
TI New pure thrombopoietin free of low-mol.weight degradation products;
purification from cell culture supernatant or milk by MPL receptor ligand-binding domain affinity chromatography and anion-exchange chromatography
AN 1996-11056 BIOTECHDS
AB A new purified mammal thrombopoietin (TPO) has a mol.weight of

70,000 +/- 10,000 (denaturing SDS-PAGE), is at least 90% pure as determined by SDS-PAGE and silver staining, and is free of TPO species of mol.weight less than 55,000. The TPO may be of mouse, primate or human origin, with a specified protein sequence. The TPO is purified from a conditioned cell culture supernatant or milk by an optional concentration step, affinity chromatography against a ligand-binding domain of an MPL receptor on crosslinked agarose beads, and anion-exchange chromatography.

TPO stimulates megakaryocytopoiesis and thrombocytopoiesis, and may be used to increase the level of platelets in the blood, e.g. in cases of aplastic anemia, myelodysplastic syndrome, chemotherapy, congenital cytopenia, etc., and may also be used to increase the number of circulating erythrocytes (or precursors), especially in therapy of anemia associated with bone marrow failure. The new TPO preparation is homogeneous and free of proteolytic degradation products. Its use reduces the need for transfusion and thus the risk of platelet alloimmunity. (91pp)

ACCESSION NUMBER: 1996-11056 BIOTECHDS
TITLE: New pure thrombopoietin free of low-mol.weight degradation products; purification from cell culture supernatant or milk by MPL receptor ligand-binding domain affinity chromatography and anion-exchange chromatography
AUTHOR: Forstrom J W; Lofton-Day C E; Lok S
PATENT ASSIGNEE: Zymogenetics
LOCATION: Seattle, WA, USA.
PATENT INFO: WO 9620955 11 Jul 1996
APPLICATION INFO: WO 1995-US16626 20 Dec 1995
PRIORITY INFO: US 1994-366859 30 Dec 1994
DOCUMENT TYPE: Patent
LANGUAGE: English
OTHER SOURCE: WPI: 1996-333942 [33]

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